



09993966-022702

Atty. Dkt. No. 014024-0280733

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Michael Rohan

Title: Human and Non-Human Primate Homologues of Nkd Protein, Nucleic Acid
Sequences Encoding, and Uses Thereof

Appl. No. 09/993,966

Filing Date: November 27, 2001

Examiner: NYA

Art Unit: 1646

RECEIVED
AUG 30 2002
TECH CENTER 1600/2900

AMENDMENT IN RESPONSE TO NOTICE UNDER 37 CFR §§1.821-825

Commissioner for Patents
Washington, D.C. 20231

Sir:

In response to the Notice to File Missing Parts of Nonprovisional Application mailed March 27, 2002, please amend the application as follows:

In the Specification:

Please amend the specification as shown:

Please delete the paragraph [0172] and replace it with the following paragraph:

[0172] The entire gene including the 5' and was cloned by the 5' RACE protocol using as the cDNA template for RACE cDNA synthesized from the total RNA obtained by LCM from a colon cancer patient as described above (RNA prep ID# 100/sample 1b3521 sample Name UC-C2CA) according to the manufacturers protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1) which is described below:

08/29/2002 AOSMAN1 00000046 033975 09993966

01 FC:117 920.00 CH

5' RACE protocol:

cDNA template for RACE was synthesized from RNA isolated by LCM from a cancer patient (RNA prep ID#100/Sample ID352/Sample name UC-C2CA) according to manufacturer's protocol (Clontech SMART RACE

cDNA Amplification Kit, K1811-1). Amplification was performed with Clontech Advantage GC-cDNA PCR kit (K1907-1) at 1M GC according to manufacturer's protocol, using cDNA template above, universal primer mix provided by manufacturer and a primer specific for human Nkd (CH308:

CTTGCCGTTGTTGTCAAAGTC) (SEQ ID NO: 23). PCR was carried out for 30 cycles of 94°C, 0.5 min/58°C, 0.5 min/68°C 2 min, followed by 10 cycles of 94°C, 1 min/58°C, 1 min/68°C 2 min. A final round of extension was carried out at 72°C for 10 min. The PCR products were cloned into pCR-TOPO4 (Invitrogen) and transformed into E. coli. Bacterial colonies harboring the correct 5'RACE product were identified by PCR screening using nested primers (CH306:

CCCAGCATGGGGAAACTTCA (SEQ ID NO: 24) and CH308:

CTTGCCGTTGTTGTCAAAGTC) (SEQ ID NO: 23).

Please delete the paragraph [0205] and replace it with the following paragraph:

[0205] In order to confirm the involvement of the hNkd gene in certain cancers, particularly colon cancer, a colon cancer cell line, SW620, was treated with β -catenin RC/AS oligos. Specifically, SC620 cells were tested with the following antisense oligos, CHIR30-5AS 5'-ACTCAGCTTGGTTAGTGTGTCAGGC-3', (SEQ ID NO: 25) and the reverse control oligos (CHIR30-5RC 5'-CGGACTGTGTGATTGGTTCGACTCA-3') (SEQ ID NO: 26). This experiment is described as follows.

REMARKS

Applicant believes that the present application is now in condition for allowance.
Favorable reconsideration of the application as amended is respectfully requested.

The Examiner is invited to contact the undersigned by telephone if it is felt that a telephone interview would advance the prosecution of the present application.

Respectfully submitted,

Date August 27, 2002

Pillsbury Winthrop, LLP
1600 Tysons Boulevard
McLean, VA 22102
Telephone: 703-905-2000
Facsimile: 703-905-2500

By 
Samir Elamrani

Attorney for Applicant
Registration No. 43,601

Should additional fees be necessary in connection with the filing of this paper, or if a petition for extension of time is required for timely acceptance of same, the Commissioner is hereby authorized to charge Deposit Account No 03-3975 for any such fees; and applicant hereby petitions for any needed extension of time.

MARKED UP VERSION ATTACHED TO AMENDMENT IN

SERIAL NO. 09/993,966

Marked up version of paragraph [0172] is below:

[0172] The entire gene including the 5' and was cloned by the 5' RACE protocol using as the cDNA template for RACE cDNA synthesized from the total RNA obtained by LCM from a colon cancer patient as described above (RNA prep ID# 100/sample 1b3521 sample Name UC-C2CA) according to the manufacturers protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1) which is described below:

5' RACE protocol:

cDNA template for RACE was synthesized from RNA isolated by LCM from a cancer patient (RNA prep ID#100/Sample ID352/Sample name UC-C2CA) according to manufacturer's protocol (Clontech SMART RACE cDNA Amplification Kit, K1811-1). Amplification was performed with Clontech Advantage GC-cDNA PCR kit (K1907-1) at 1M GC according to manufacturer's protocol, using cDNA template above, universal primer mix provided by manufacturer and a primer specific for human Nkd (CH308:

CTTGCCGTTGTTGTCAAAGTC) (**SEQ ID NO: 23**). PCR was carried out for 30 cycles of 94°C, 0.5 min/58°C, 0.5 min/68°C 2 min, followed by 10 cycles of 94°C, 1 min/58°C, 1 min/68°C 2 min. A final round of extension was carried out at 72°C for 10 min. The PCR products were cloned into pCR-TOPO4 (Invitrogen) and transformed into E. coli. Bacterial colonies harboring the correct 5'RACE product were identified by PCR screening using nested primers (CH306:

CCCAGCATGGGGAAACTTCA (**SEQ ID NO: 24**) and CH308:

CTTGCCGTTGTTGTCAAAGTC) (**SEQ ID NO: 23**).

Atty. Dkt. No. 014024-0280733

Marked up version of paragraph [0205] is below:

[0205] In order to confirm the involvement of the hNkd gene in certain cancers, particularly colon cancer, a colon cancer cell line, SW620, was treated with β -catenin RC/AS oligos. Specifically, SC620 cells were tested with the following antisense oligos, CHIR30-5AS 5'-ACTCAGCTTGGTTAGTGTGTCAGGC-3', (**SEQ ID NO: 25**) and the reverse control oligos (CHIR30-5RC 5'-CGGACTGTGTGATTGGTTCGACTCA-3') (**SEQ ID NO: 26**). This experiment is described as follows.